

Examination of Heart Rate and Intramuscular Oxygen Saturation Variability in the Gradually Increasing Running Test

Miray İLHAN¹, Serdar Orkun PELVAN²

Abstract

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Aim: This study aims to examine physiological responses during a gradually increasing running test by simultaneously evaluating heart rate (HR), heart rate variability (HRV), and muscle oxygen saturation (SmO₂) parameters

Materials and Methods: Sixteen male athletes aged between 18 and 27 years, with at least 3 years of sports experience, participated in the study. The average age was 19.25 ± 2.21 years, average height was 180 ± 7.87 cm, average weight was 78.3 ± 14.6 kg, and average body fat percentage was 11.8 ± 5.85 . HR, HRV, and SmO₂ values were recorded during the resting, running, and recovery phases throughout the test. The running protocol started at a speed of 10 km/h and increased by 1 km/h every minute until exhaustion. Recording continued for 5 minutes post-run.

Results: During the gradually increasing running test, it was observed that heart rate (HR) increased while SmO₂ levels decreased. Throughout the recovery period after running, both measurement parameters tended to approach resting values

Conclusion: These findings indicate that the joint assessment of HR, HRV, and SmO₂ is significant for a holistic analysis of the physiological responses to exercise. In performance evaluation, measurements such as heart rate (HR), heart rate variability (HRV), and intramuscular oxygen saturation (SmO₂) hold an important place alongside running tests.

Kademeli Artan Koşu Testinde Kalp Atım Hızı ve Kas İçi Oksijen Saturasyon Değişkenliğinin İncelenmesi

Özet

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Amaç: Bu çalışma, kalp atım hızı (HR), kalp atım hızı değişkenliği (HRV) ve kas içi oksijen saturasyonu (SmO₂) parametrelerini birlikte değerlendirerek, kademeli artan koşu testi sırasında fizyolojik tepkileri incelemeyi amaçlamaktadır.

Gereç ve Yöntem: Çalışmaya, 18–27 yaş aralığında, en az 3 yıllık spor geçmişine sahip, yaş ortalaması 19,3 yıl, boy ortalaması $180 \pm 7,87$ cm, kilo ortalaması $78,3 \pm 14,6$ kg ve vücut yağ oranı ortalaması $11,8 \pm 5,85$ olan 16 erkek sporcu katılmıştır. Dinlenme, koşu ve toparlanma evrelerinde HR, HRV ve SmO₂ değerleri test süresince kaydedilmiştir. Koşu protokolü, 10 km/h hızla başlatılmış ve her dakika 1 km/h artırılarak tükenişe kadar sürdürülmüştür. Koşu sonrası 5 dakika boyunca kayıt alınmaya devam edilmiştir.

Bulgular: Bulgular: Kademeli artan koşu testi sırasında HR'nin arttığı, SmO₂ seviyelerinin ise azaldığı gözlemlenmiştir. Koşu sonrası toparlanma süresince, her iki ölçüm parametresi de dinlenik değerlere yaklaşma eğilimi göstermiştir.

Sonuç: Bu bulgular, HR, HRV ve SmO₂'nin birlikte değerlendirilmesinin egzersize verilen fizyolojik yanıtların bütüncül analizinde önemli olduğunu göstermektedir. Performans değerlendirmede koşu testleri ile birlikte kalp atım hızı (HR) ve değişkenliği (HRV), kas içi oksijen saturasyonu (SmO₂) gibi ölçümler önemli bir yer tutmaktadır.

¹ Marmara Üniversitesi / Sağlık Bilimleri Enstitüsü, Beden Eğitimi ve Spor Anabilim Dalı/ İstanbul Türkiye / mryilhan@gmail.com ORCID: 0000-0003-1404-7902

² Marmara Üniversitesi/Spor Bilimleri Fakültesi, Spor Sağlık Bilimleri Anabilim Dalı/ İstanbul, Türkiye / orkunpelvan@gmail.com ORCID: 0000-0001-7809-0788

Introduction

The physiology of running is a widely researched topic aimed at examining the physiological responses of the human body during exercise (Cassirame et al., 2022). Gradually increasing running tests are frequently used to evaluate these physiological responses. Particularly, the gradual increase in exercise intensity allows for the determination of key parameters in assessing aerobic and anaerobic performance

(Carminatti et al., 2013). In these tests, variables such as speed or incline can be adjusted for each stage (Bentley et al., 2007).

In performance evaluation, measurements such as heart rate (HR) and its variability (HRV), along with muscle oxygen saturation (SmO_2), play an important role in running tests. HR refers to the total number of heartbeats in one minute. The concept of HRV refers to the variations in the duration between each heartbeat (McCraty, Shaffer, 2015). SmO_2 is a parameter measured by near-infrared spectroscopy (NIRS), representing the ratio of oxygen-carrying hemoglobin in muscle tissue to the total hemoglobin (Perrey et al., 2024). This value indicates the balance between the oxygen reaching the muscles and the level at which the muscle uses this oxygen (Quaresima, Ferrari, 2002).

Maximal load exercises performed at a constant load are applied at a specific intensity without changing the intensity. In such exercises, SmO_2 levels may initially remain stable but show a decrease over time (Perrey, S., & Rupp, T. 2009). Unlike these exercises, where muscles provide more predictable responses due to continuous oxygen demand, gradually increasing running tests offer a more comprehensive evaluation of individuals' maximum oxygen consumption and HR. While HRV reflects the functions of the autonomic nervous system, SmO_2 emerges as a critical parameter for assessing the aerobic capacity and oxygen utilization of muscles. In gradually increasing running tests, as exercise intensity increases, a noticeable decrease in SmO_2 levels is observed, along with a parallel increase in heart rate (Kalkan, Pelvan, 2023). Therefore, this study employs a gradually increasing running test to understand how heart rate and muscle oxygen levels change in relation to increased running performance.

It is suggested that the decrease in muscle oxygen levels during exercise is closely related to the responses of the circulatory system (Grassi et al., 2003). During the recovery process post-exercise, HRV is directly related to the re-engagement of the parasympathetic nervous system, and various studies have been conducted on the role of SmO_2 in this process (Stanley et al., 2013). These findings reveal the importance of these parameters in understanding the complex relationship between SmO_2 , HR, and HRV during gradually increasing running tests and the physiological adaptations of the body (Stanley et al., 2013). Especially the RR values measured during the recovery phase help to better understand the effects of different exercise models on the cardiovascular system (Kalkan, Pelvan, 2023). Findings in the literature have examined the effects of acute fatigue on HRV and SmO_2 , concluding that fatigue increases sympathetic nervous system activity by enhancing muscle oxygen consumption, while reducing HRV (Perrey, S., & Rupp, T. 2009). These findings highlight that physiological responses related to exercise intensity and duration have significant effects on the nervous system and muscle metabolism.

This study aims to investigate the body's responses during a gradually increasing running tests by jointly evaluating physiological parameters such as HR, HRV, and SmO_2 . It aims to examine the effects of graded exercise on cardiovascular and muscle oxygenation by monitoring resting HR and SmO_2 with HRV during the post-exercise recovery process. Previous studies have typically examined heart rate variability (HRV) and muscle oxygen saturation (SmO_2) separately during exercise. However, there is a lack of studies that simultaneously evaluate these parameters throughout a graded running protocol, including both exercise and recovery phases. This study aims to address this gap by jointly assessing HR, HRV, and SmO_2 to provide

an integrated understanding of cardiovascular and muscular responses. We hypothesize that graded exercise will lead to a progressive increase in HR and a decrease in HRV and SmO_2 , with recovery patterns varying depending on the physiological parameter measured. We hypothesize that graded exercise will cause significant changes in HR, HRV, and SmO_2 , and that the type of recovery protocol will influence the rate and extent of cardiovascular and muscle oxygenation recovery.

Materials And Methods

Study Group

Athletes aged 18-35, who are eligible to compete in the Türkiye Championship and have at least 3 years of licensed sports experience without any health issues, comprised the participant group for this study. The athletes participating in the research voluntarily joined the study in accordance with the World Medical Association (WMA) Declaration of Helsinki. Ethical approval was obtained from the Clinical Research Ethics Committee of Marmara University Faculty of Medicine with protocol number 09.2024.527.

Data Collection Tools

Height and Body Weight

The participants' body weights, body fat percentages, lean body weights, and body mass indices were measured using a 4-probe TANITA brand MC 780 MA model Bioelectrical Impedance device. Prior to the measurement, participants were informed to ensure the reliability of the measurements.

Heart Rate Data Acquisition

Throughout the measurement period, HR and HRV values were recorded before, during, and after the test using a chest strap Polar H10 heart rate monitor to observe HR and HRV. The obtained data were evaluated separately as pre-run (resting), running test (loading), and recovery.

Near-Infrared Spectroscopy (NIRS) for SmO_2 Measurement

Biological tissues possess a structure that allows the penetration of light. This property enables light to propagate through the tissue and reach nearby photoreceptors. During its passage through the tissue, light is partially absorbed by hemoglobin molecules. The amount of light detected is particularly influenced by changes in the concentrations of hemoglobin and oxyhemoglobin in the blood. This method employs the near-infrared (NIR) light spectrum in the range of 700–1000 nm, a wavelength range characterized by high transmittance in biological tissues. To measure this physiological process, a specialized device is placed on the skin. This device functions both as a light source and as a detector of the reflected light intensity. Infrared light emitted from the device propagates through the tissue and reaches nearby photoreceptors. Variations in the intensity of these light signals enable the collection of information regarding oxyhemoglobin and deoxyhemoglobin levels at the tissue surface. SmO_2 measurements of the participants were conducted using a wireless and portable TRAINRED model near-infrared spectroscopy (NIRS) device. For the measurement, the device was placed on the lateral part of the M. Vastus Lateralis muscle, starting from the lateral epicondyle of the patella, and 15 cm above, using a measuring tape. To maintain measurement accuracy, care was taken to ensure that the device did not come over prominent vascular structures.

Additionally, the device was isolated from external factors such as light and sweat to ensure accurate and reliable results, aiming to conduct the measurement process under optimal conditions.

Experimental Design

The target population of the research consists of 16 active and licensed participants aged 18-35. Participants' heart rates were monitored using a Polar H10 chest strap heart rate monitor, and resting heart rates were recorded in a supine position, isolated from sound and light, for 10 minutes. In the measurement of heart rate and intramuscular oxygen saturation taken during a ten-minute resting period, the first three minutes and the last three minutes of data were disregarded to observe the participant's resting state (intense resting phase). Maximum, minimum, and average HR, HRV, and SmO₂ values were noted during the 4th, 5th, 6th, and 7th minutes of the measurement. At the end of the rest, participants were given 10 minutes for general and specific warm-ups. Upon completion of the warm-up, information about the gradually increasing running tests was provided, and a 5-minute rest was given before the test. SmO₂, HR, and HRV before starting the run were noted. The test applied to participants on the treadmill was set to start at 10 km/h and increase by 1 km/h every minute. SmO₂, HR, and HRV were continuously recorded throughout the measurement. When the test began, device recording was initiated, and the maximum duration and speed values obtained by the participants were recorded by applying the gradually increasing running tests on the treadmill. Immediately after the gradually increasing running tests ended, participants were asked to lie down in a supine position again, and 5-minute recovery data were recorded under sound and light isolation.

Data Analysis

Normalization of Data

The running test continued until the participant reached a level where they could no longer continue. Therefore, the running time varied for each participant. Due to the different running times, the data for each athlete was divided into ten equal parts for normalization. Thus, each value was compared both within itself and with other participants.

SmO₂ Analysis

All data were transferred to a computer. Data were organized for signal analysis using the Procalysis program. For statistical analysis, the Jamovi (version 2.5.3) program was used. In the 10-minute resting muscle oxygen saturation measurement, the averages of the maximum, minimum, and average muscle oxygen saturation values during the 4th, 5th, 6th, and 7th minutes were noted. Variations within each stage over time were compared. During the recovery process, data were divided into 30-second periods and analyzed as maximum, minimum, and average values. The obtained data were analyzed using Repeated Measures ANOVA test in the Jamovi program. A significance level of $p < 0.05$ was accepted for statistical analyses.

HR and RR Analysis

After the measurements were completed, the first analysis of the data was performed using the Kubios HRV standard 3.5.0 program. Maximum, minimum, average HR, and average RR values were taken. At the end of the run, a five-minute recovery data was recorded for the measurement of resting heart rate in the participants. These data were divided into 30-second periods and analyzed as maximum, minimum,

average HR, and average RR values. For statistical analysis, the Jamovi (version 2.5.3) program was used. To evaluate changes at different performance levels, a Repeated Measures ANOVA test was conducted, and a significance level of $p < 0.05$ was accepted while examining whether there was a significant difference between running stages.

Results

Descriptive characteristics and running speeds of the 16 athletes participating in this study are presented in Table 1.

Table 1. Descriptive characteristics of the participants and the maximal running speed achieved during the test.

	N	Minimum	Maksimum	\bar{x}	SS
Height (cm)	16	165	193	180	7.87
Weight (kg)	16	58.5	99.9	78.3	14.6
Age	16	18	27	19.3	2.21
Fat Rate (%)	16	3.1	22.9	11.8	5.85
Maximal Running Speed (km/h)	16	16	23	17.6	2.03

Gradually Increasing Running Tests Analyses

The Post Hoc analysis determined that the sphericity assumption was valid. A statistically significant increase in HR values was observed with each speed increase during the test ($p < .001$ for all stages; $p = .018$ for the last stage). Table 2 shows the changes in heart rate levels of participants during the gradually increasing running tests for each stage separately.

Table 2. Changes in heart rate (HR) during the incremental running test

HR Factor 1	HR Factor 1	Mean Difference	SE	df	t	p _{tukey}
REST*	- Pre- Test*	-38.30	4.163	15.0	-9.20	< .001
Pre-Test	- Level 1	-24.79	2.920	15.0	-8.49	< .001
Level 1	- Level 2	-23.91	2.989	15.0	-8.00	< .001
Level 2	- Level 3	-9.13	1.004	15.0	-9.09	< .001
Level 3	- Level 4	-6.87	0.564	15.0	-12.19	< .001
Level 4	- Level 5	-5.73	0.585	15.0	-9.80	< .001
Level 5	- Level 6	-4.41	0.426	15.0	-10.37	< .001
Level 6	- Level 7	-3.77	0.360	15.0	-10.47	< .001
Level 7	- Level 8	-3.60	0.270	15.0	-13.35	< .001
Level 8	- Level 9	-3.45	0.218	15.0	-15.81	< .001
Level 9	- Level 10	-2.32	0.529	15.0	-4.39	0.018

Rest: Average HR values of participants during the 4-minute resting period

Pre-Test: HR value immediately before the onset of running

A statistically significant difference was observed in RR intervals across all stages during the running test ($p < .001$ for the first 10 stages, $p = .026$ for the last stage). A decrease in RR value was observed with the increase in running levels (Table 3). The average HR-RR changes of participants during the gradually increasing running tests are presented in Figure 1.

Table 3. Changes in respiratory rate (RR) during the incremental running test

RR Factor 1	RR Factor 1	Mean Difference	SE	t	p _{tukey}	Cohen's d
REST*	- Pre-Test*	887.19	38.276	23.18	< .001	5.79
Pre-Test	- Level 1	-452.48	11.920	-37.96	< .001	-9.49
Level 1	- Level 2	63.77	4.958	12.86	< .001	3.22
Level 2	- Level 3	22.66	2.760	8.21	< .001	2.05
Level 3	- Level 4	15.08	1.509	9.99	< .001	2.50
Level 4	- Level 5	11.66	1.348	8.65	< .001	2.16
Level 5	- Level 6	8.50	0.992	8.57	< .001	2.14
Level 6	- Level 7	6.90	0.766	9.01	< .001	2.25
Level 7	- Level 8	6.25	0.508	12.31	< .001	3.08
Level 8	- Level 9	5.73	0.384	14.93	< .001	3.73
Level 9	- Level 10	3.70	0.885	4.18	0.026	1.05

Rest: Average HR values of participants during the 4-minute resting period Pre-Test: HR value immediately before the onset of running

The average changes in heart rate (HR) and respiratory rate (RR) of the participants during the incremental running test are presented in Figure 1.

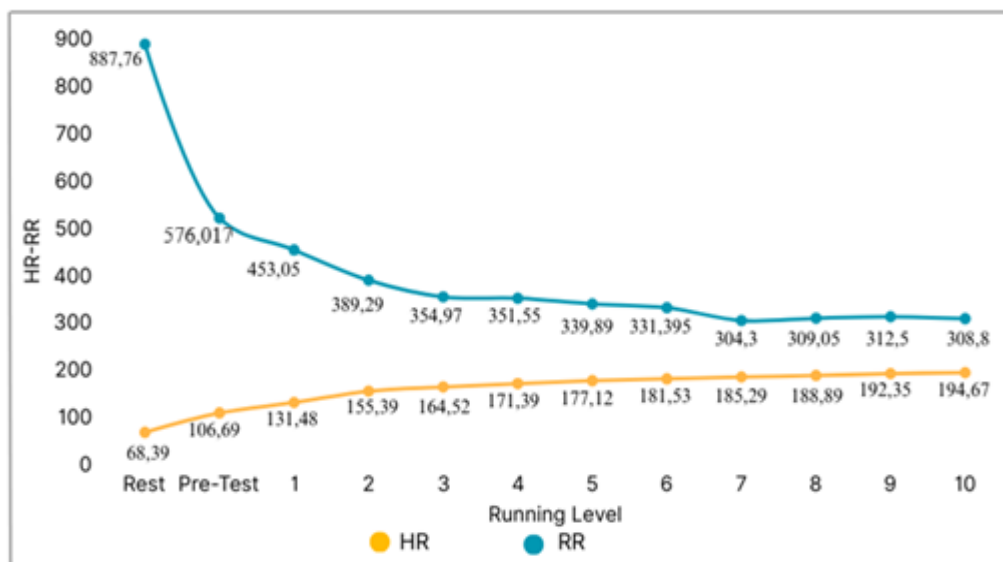


Figure 1. HR-RR Change Graph

When SmO₂ data were examined, no significant difference was found between REST and Pre-Test ($p=1.000$), but a statistically significant decrease was detected at the beginning of the test ($p<.001$). No significant change was observed in SmO₂ levels between the second and eighth stages ($p>.05$). A significant decrease was observed again before the last stage ($p = .010$), but no statistically significant difference was found in the final stage of the run ($p = .745$). The change in SmO₂ with increasing running levels is presented in Table 4.

Table 4. Changes in muscle oxygen saturation (SmO₂) during the incremental running test

SMO ₂ Factor 1	SMO ₂ Factor 1	Mean Difference	SE	df	t	p _{tukey}
REST*	- Pre-Test*	1.084	1.497	15.0	0.724	1.000
Pre-Test	- Level 1	3.735	0.471	15.0	7.928	<.001
Level 1	- Level 2	3.341	0.730	15.0	4.575	0.013
Level 2	- Level 3	1.343	0.431	15.0	3.113	0.167
Level 3	- Level 4	1.049	0.350	15.0	2.998	0.200
Level 4	- Level 5	0.923	0.327	15.0	2.822	0.261
Level 5	- Level 6	0.240	0.538	15.0	0.446	1.000
Level 6	- Level 7	1.310	0.408	15.0	3.212	0.142
Level 7	- Level 8	1.485	0.459	15.0	3.237	0.136
Level 8	- Level 9	2.437	0.515	15.0	4.732	0.010
Level 9	- Level 10	2.744	1.446	15.0	1.898	0.745

Rest: Average HR values of participants during the 4-minute resting period

Pre-Test: HR value immediately before the onset of running

The changes in muscle oxygen saturation (SmO₂) of the participants during the incremental running test are presented in Figure 2.

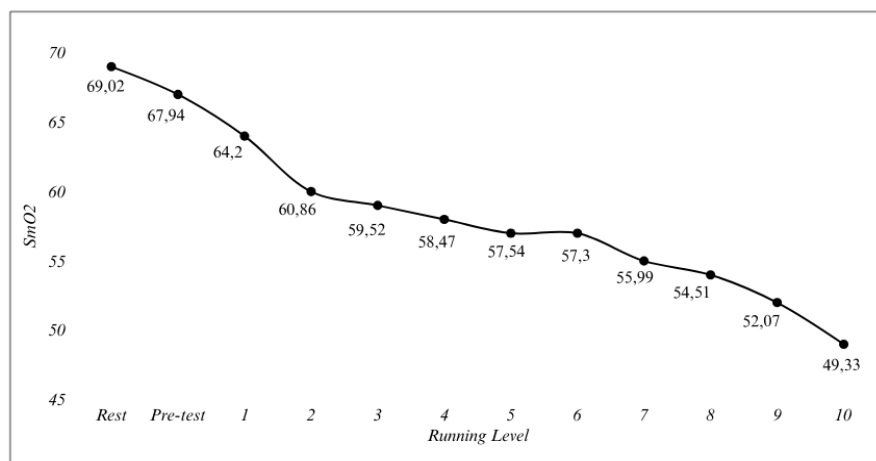


Figure 2. SmO₂ Change Graph

Analysis of Recovery Data Following the Incremental Running Test

During the post-exercise recovery period, a statistically significant decrease in heart rate (HR) was observed within the first 2.5 minutes ($p < .001$, $p = .015$). However, no significance was observed in HR values from the 180th second onward ($p > .05$). The changes in participants' heart rate throughout the recovery period for each time interval are presented separately in Table 5.

Table 5. Changes in heart rate (HR) following the incremental running test

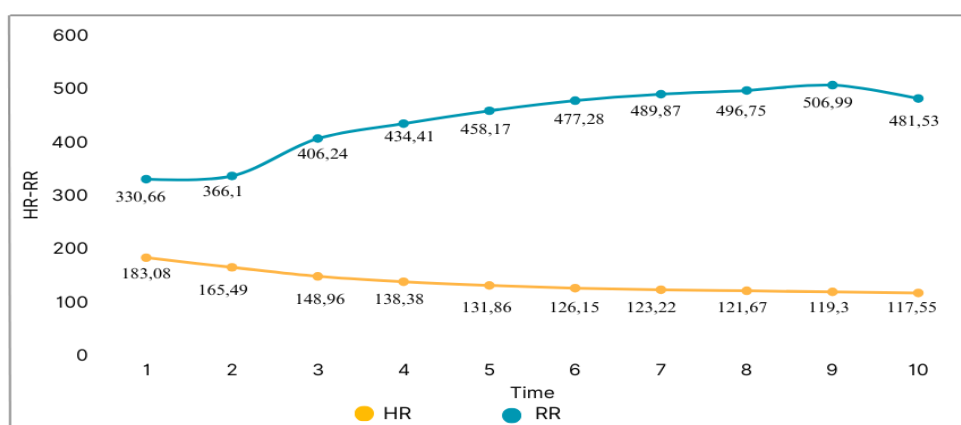
HR Factor 1		HR Factor 1	Mean Difference	SE	df	t	p _{Tukey}
Level 1	-	Level 2	17.59	1.491	15.0	11.79	< .001
Level 2	-	Level 3	16.54	1.336	15.0	12.38	< .001
Level 3	-	Level 4	10.58	1.187	15.0	8.91	< .001
Level 4	-	Level 5	6.52	0.709	15.0	9.21	< .001
Level 5	-	Level 6	5.71	0.836	15.0	6.83	< .001
Level 6	-	Level 7	2.93	0.676	15.0	4.33	0.015
Level 7	-	Level 8	1.55	0.555	15.0	2.79	0.224
Level 8	-	Level 9	2.37	0.745	15.0	3.19	0.118
Level 9	-	Level 10	1.75	1.047	15.0	1.67	0.797

RR intervals significantly decreased with the onset of rest ($p<.05$). No statistically significant differences were found when examining 210-240 seconds ($p=.194$) and at the end of the rest ($p=.998$). The RR change of participants during the recovery period is presented in Table 6.

Table 6. Changes in respiratory rate (RR) following the incremental running test

RM Factor 1		RM Factor 1	Mean Difference	SE	df	t	p _{Tukey}
Level 1	-	Level 2	-35.44	3.36	15.0	-10.552	<.001
Level 2	-	Level 3	-40.14	3.15	15.0	-12.744	<.001
Level 3	-	Level 4	-28.17	4.62	15.0	-6.098	<.001
Level 4	-	Level 5	-23.76	3.15	15.0	-7.539	<.001
Level 5	-	Level 6	-19.11	2.97	15.0	-6.441	<.001
Level 6	-	Level 7	-12.60	2.24	15.0	-5.612	0.001
Level 7	-	Level 8	-6.88	2.39	15.0	-2.881	0.194
Level 8	-	Level 9	-10.23	2.76	15.0	-3.706	0.047
Level 9	-	Level 10	25.46	32.92	15.0	0.773	0.998

The average changes in heart rate (HR) and respiratory rate (RR) of the participants after the incremental running test are presented in Figure 3.

*Figure 3. Post-exercise HR-RR Change*

During the recovery period after the run, a significant increase in SmO₂ values was observed in the first 30 seconds ($p < .001$). A statistically significant change was also detected between 30–60 seconds ($p = .033$). However, no significant change in SmO₂ levels was observed from the 60th second onwards ($p > .05$). No difference was found between 120–210 seconds ($p = 1.000$).

Table 7. Changes in muscle oxygen saturation (SmO₂) following the incremental running test

SMO ₂ Factor 1		SMO ₂ Factor 1	Mean Difference	SE	df	t	p _{Tukey}
Level 1	-	Level 2	-7.93888	1.026	15.0	-7.74011	< .001
Level 2	-	Level 3	-5.77371	1.479	15.0	-3.90275	0.033
Level 3	-	Level 4	-2.76261	0.931	15.0	-2.96803	0.169
Level 4	-	Level 5	-1.02602	0.353	15.0	-2.90544	0.186
Level 5	-	Level 6	0.34584	0.666	15.0	0.51964	1.000
Level 6	-	Level 7	-0.10455	0.496	15.0	-0.21060	1.000
Level 7	-	Level 8	-0.00263	0.486	15.0	-0.00541	1.000
Level 8	-	Level 9	0.49493	0.245	15.0	2.02142	0.599
Level 9	-	Level 10	-0.43951	0.527	15.0	-0.83367	0.997

The changes in muscle oxygen saturation (SmO₂) of the participants after the incremental running test are presented in Figure 4.

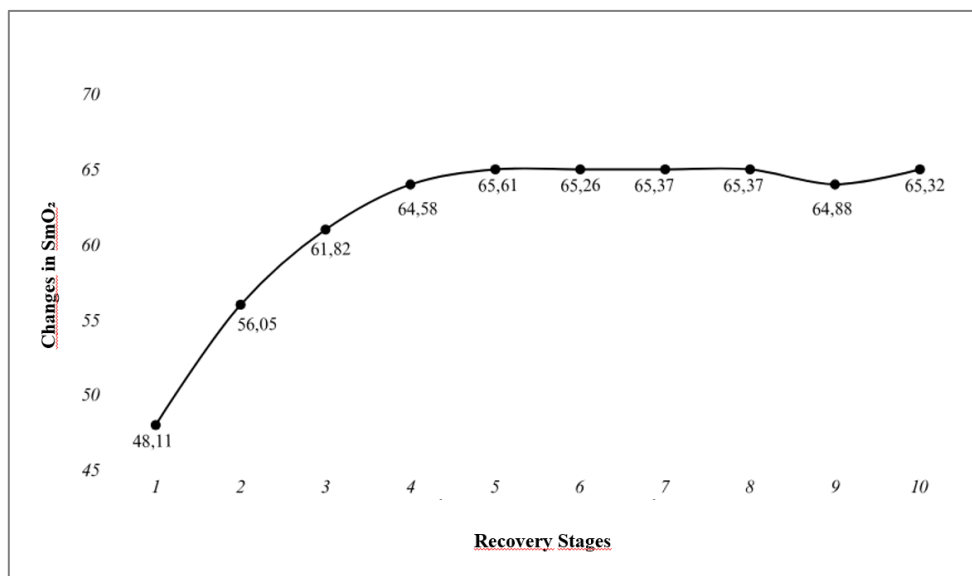


Figure 4. Post-exercise SmO₂ Change

Discussion and Conclusion

The HR, HRV, and SmO₂ data obtained in the study reveal the dynamic responses exhibited by the organism as exercise intensity increases, approaching physiological limits. In the literature, there are studies measuring SmO₂ in gradually increasing running tests (Pelvan et al., 2024), (Kalkan, Pelvan, 2023). However, studies evaluating the recovery process after running and SmO₂, HR, and HRV together have not been encountered. In many studies related to SmO₂, it is observed that the groups included in the study generally consist of athletes with low training levels or patients.

In the graded running test, a significant increase in HR values was observed parallel to the increase in exercise intensity, while a decrease occurred in RR values (Figure 1). Changes in HRV show that the heart does not always work like a metronome and has a dynamic structure. The significant increase in HR during exercise can be explained by the sympathetic nervous system becoming dominant, resulting in increased cardiac output due to the interaction between the parasympathetic and sympathetic nervous systems. In a resting state, the parasympathetic nervous system (PNS) is active, trying to keep the heart rate between 60-80 bpm (Poehling, Llewellyn, 2019). This situation indicates that the cardiovascular system works more intensively to meet increasing metabolic demands.

During the running test, a decrease in HRV was observed as the running stage increased. This may be due to a reduction in vagal modulation to the heart and an increase in sympathetic nerve activity (Tulppo et al., 1997). Changes in HRV can stabilize at a certain point, limiting the variability between heartbeats. As fatigue develops, the increase in HR may slow down or reach plateau levels. This situation may indicate that the individual is approaching their maximal aerobic capacity or that central fatigue processes are coming into play (Midgley et al., 2007). HR data show a parallel increase with sensitivity to exercise intensity. While HR significantly increases with each stage of the test ($p < .001$), the statistical significance level in the final stage is relatively lower ($p = .018$), which may be attributed to individual maximal response differences, exercise fatigue, or the participant approaching their maximal HR level (Table 2). This linear increase trend indicates that the cardiovascular system is becoming increasingly activated in accordance with the physiological loading structure of the test (Achten, Jeukendrup, 2003). The increase in HR can be explained by the manifestation of sympathetic activity and the suppression of parasympathetic activity. This situation is consistent with the systemic cardiovascular response to exercise loading. In a progressively increasing running test, the increase in exercise intensity every minute leads to significant changes in autonomic nervous system activity to meet the organism's metabolic needs. The increase in heart rate before running is primarily explained by the withdrawal of parasympathetic tone, while as exercise intensity increases, it may be attributed to the prominence of sympathetic stimulation (Michael et al., 2017). Increased sympathetic activity triggers cardiovascular responses such as heart rate, myocardial contractility, and peripheral vasoconstriction, thereby facilitating the delivery of more oxygen to the muscles in response to the increased oxygen demand (Rowell, 1993). At the same time, catecholamines released from the adrenal medulla (especially epinephrine and norepinephrine) enhance these physiological responses, increasing sympathetic effectiveness (Powers & Howley, 2017). Blood flow to the muscles increases during exercise, achieved through local vasodilation in the working muscles, while sympathetic vasoconstriction becomes dominant in non-working areas (Joyner & Casey, 2015). These findings indicate that sympathetic activity plays a central role in the physiological adaptation process in the later stages of the test.

SmO₂ is used in performance and metabolic stress evaluations as it reflects the balance between the level of oxygen delivered to peripheral muscles and consumption (Van Beekvelt et al., 2001). In SmO₂ values, a more pronounced percentage decrease occurred especially between the first and second stages, while this decrease occurred at lower levels between subsequent stages (Figure 2). This situation may be related to the participants directly transitioning to running pace due to the test starting at a speed of 10 km/h.

The decrease in SmO_2 during exercise may lead to increased activity of the sympathetic nervous system, altering the cardiovascular response. Consequently, an increase in HR levels may have been observed. This mechanism is regulated through chemosensitive receptors and metabolic reflexes (Mitchell et al., 2019).

The decrease in SmO_2 levels observed during running may be due to the activation of oxidative capacity to meet the increasing energy demands. While no significant variability in SmO_2 levels was observed throughout the progressively increasing running test (Table 4), a marked increase in HR occurred. This situation may be attributed to the cardiovascular system responding to the increase in exercise intensity, thereby helping to maintain more stable local muscle oxygenation. One possible reason for this stability in SmO_2 levels is the ability of the muscle groups used to maintain the balance of oxygen utilization and recovery during exercise. On the other hand, the continuous increase in HR values may be related to the rise in sympathetic nervous system activation and the increase in metabolic demand (Achten & Jeukendrup, 2003). As exercise progresses, the heart needs to work more both to transport oxygen-rich blood and to regulate heat, which may explain the increase in HR. Peripheral (muscle-level) physiological responses may not always progress parallel with central (cardiovascular) system responses; this reveals the specificity of different physiological systems in their responses to exercise. A significant decrease in SmO_2 levels was observed before running and in the first two stages of the test ($p < .001$, $p = .013$), indicating that oxygen consumption increased to meet the muscle's increased oxygen demand (Table 4). As mentioned in the literature, muscle oxygen saturation decreases with increasing exercise intensity, and this situation is associated with local oxygen increase (McManus et al., 2018). However, no statistically significant change was detected between the 2nd and 8th stages ($p > .05$). This may suggest that the muscle has reached a temporary oxygen balance state or that peripheral circulation has adapted to exercise, stabilizing oxygen delivery. A significant decrease was observed again in the 9th stage ($p = .010$), but this significance disappeared in the final stage ($p = .745$). These findings show that SmO_2 response may not follow a linear decrease pattern during exercise and may reach less variable decrease levels due to factors such as individual tolerance differences, intramuscular metabolic responses, or circulatory system adaptations.

During the recovery process after running, the variability of SmO_2 , HR, and HRV parameters at different time intervals is due to these physiological indicators being regulated by different systems. In this study, significant recovery was observed in SmO_2 values only in the first 60 seconds, while no significant variability was observed in the following 4-minute period (Table 6). In contrast, HR and HRV parameters continued to show significant changes throughout recovery, especially up to the first 180 seconds (Table 5). This difference may be due to muscle oxygen saturation (SmO_2) quickly reaching metabolic balance at the local level. Increased muscle blood flow with post-exercise reactive hyperemia may rapidly increase oxygen transport, causing SmO_2 to reach normal levels in the first minute. HR and HRV may return to normal levels with the reactivation of the autonomic nervous system, especially parasympathetic tone; this process can be observed more slowly, continuing up to the first 2-3 minutes after exercise (Stanley et al., 2013). Additionally, the stability of SMO_2 shows that the balance between oxygen consumption at the muscle level and oxygen provided from circulation is quickly achieved (Perrey & Ferrari, 2018). In contrast, HRV recovery takes longer because it is affected not only by metabolic factors but also by respiratory, neural

feedback, and hormonal factors. These findings are important in showing that peripheral and central physiological systems return to normal at different time scales during the recovery process. This situation shows that the muscle's oxygenation capacity quickly recovers at the beginning of the recovery process. However, the lack of significant change in subsequent minutes suggests that SmO_2 quickly reaches plateau levels and peripheral circulation effectively recovers.

SmO_2 can increase lactate accumulation by triggering tissues to shift towards anaerobic energy production and can increase metabolic stress. This process can lead to an increase in HR, causing HRV to change towards sympathetic activity. The duration and intensity of exercise affect the extent of these changes, contributing to HR increasing significantly during high-intensity exercises but reaching a more balanced level over time. Research shows that the decrease in SmO_2 is associated with increased sympathetic nervous system activity and consequently a decrease in HRV (Perrey, S., & Rupp, T. 2009). Our study similarly reported that SmO_2 showed a rapid increase during the recovery phase and then stabilized (Boone et al., 2016). In the literature, the rate of increase in SmO_2 levels during recovery is considered a reflection of aerobic capacity and peripheral circulation efficiency. In this context, the recovery trend in our study suggests that participants have a high level of circulatory adaptation and achieve a rapid homeostatic balance after exercise. The obtained data allow for the evaluation of not only performance but also recovery capacity after exercise. The combined examination of HR and SmO_2 data provides an opportunity to comprehensively evaluate physiological loading responses at both central (cardiovascular) and peripheral (muscle) levels (Ferrari et al., 2011).

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Suggestions

- Future studies could include different age groups or genders to increase the generalizability of the findings.
- The long-term effects of recovery protocols and HRV and SmO_2 responses at varying exercise intensities could be examined. Studies with larger sample sizes and participants from diverse sports disciplines would allow a more comprehensive assessment of physiological responses during graded exercise.
- Including additional metabolic or hormonal parameters alongside muscle oxygenation measurements could provide a more holistic evaluation of exercise and recovery processes.

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